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# Four new *Phragmidium* (Phragmidiaceae, Pucciniomycetes) species from Rosaceae plants in Guizhou Province of China

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## Abstract

In this study, four new species of *Phragmidium* were proposed based on morphological and molecular characters. In morphology, *Phragmidium rosae-roxburghii* sp. nov. was distinguished to related taxa by its unique square to diamond-shaped urediniospores; *Ph. rubi-coreani* sp. nov. differed from *Ph. barclayi* and *Ph. cibanum* because of teliospores with fewer cells and shorter pedicels; urediniospores of *Ph. potentillae-freynianae* sp. nov. were bigger than *Ph. duchesneae-indica*; and *Ph. rosae-laevigatae* sp. nov. produced bigger urediniospores than *Ph. jiangxiense*. The phylogenetic analyses based on the combination of two loci (ITS and LSU) also supported our morphological conclusion. In the meantime, three previously known species were also described herein.

## Keywords

Basidiomycota, ITS, LSU, phylogeny, rust disease, taxonomy

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## Introduction

*Phragmidium* (Phragmidiaceae) was established by Link (1816) and characterized by laterally separated multicellular teliospores with pigmented bilaminar walls, and a thickened pedicel at the base (Wei 1988).

The genus was widely distributed around the world especially in the northern hemisphere, such as China, USA and Japan (Wei 1988; Zhuang 1989; Cummins and Hiratsuka 2003; Maier et al. 2003; Zhuang et al. 2012; Pscheidt and Rodriguez 2016; Liu et al. 2018, 2019, 2020; Zhao et al. 2021). *Phragmidium* species often caused severe rust diseases in Rosaceae plants (*Rosa*, *Rubus*, *Potentilla*, *Sanguisorba*, *Duchesnea* and *Acaena*). Species of *Phragmidium* have been reported growing on host plants of *Rosa*, *Rubus*, and *Potentilla*, with a few species on *Sanguisorba* (Cummins and Hiratsuka 2003; Maier et al. 2003; Yun et al. 2011; Pscheidt and Rodriguez 2016; Liu et al. 2018, 2019, 2020), *Duchesnea* (Zhao et al. 2021) and *Acaena* (McTaggart et al. 2016). Two species *Ph. mucronatum* (Pers.) Schltdl. and *Ph. tuberculatum* Jul. Müll., were common pathogens on ornamental roses worldwide (Wahyuno et al. 2001, 2002; Leen and Van Huylenbroeck 2007; Wilson and Aime 2014).

About 8000 species of rust fungi have been reported in the world (Zhao et al. 2021). Based on morphological features or host associations, 1200 species belonging to 71 genera of 15 families were previously reported in China. Over 70 *Phragmidium* species have been described (Cummins 1931; Arthur 1934; Zhuang et al. 1998, 2003, 2005, 2012; Wahyuno et al. 2001; Cummins and Hiratsuka 2003; Yang et al. 2015; Ali et al. 2017; Aime et al. 2018; Liu et al. 2018, 2019, 2020; Ono and Wahyuno 2019; Aime and McTaggart 2021; Zhao et al. 2021).

Traditionally, *Phragmidium* species are distinguished based on teliospores morphology (Wei 1988). According to Wahyuno et al. (2001) and Zhao et al. (2021) *Phragmidium* species are difficult to distinguish based only on morphology of asexual spore stages; thus, DNA data is essential for taxonomy and identification of *Phragmidium* species.

The combination of morphological and molecular characters has been applied in the taxonomy of rust fungi (Beenken et al. 2012; Beenken 2014; McTaggart et al. 2016, 2017; Liu et al. 2018, 2019, 2020; Ono and Wahyuno 2019; Zhao et al. 2021). *Phragmidium* includes more than 270 epithet records which are listed in MycoBank (https://www.mycobank.org) and Index Fungorum (http://www.indexfungorum.org) (accessed in October 2022). However, only 28 records were described and named by Chinese researchers, three *Phragmidium* taxa in Guizhou Province, *Ph. duchesneae-indicae*, *Ph. nonapiculatum* and *Ph. kans* were introduced by Zhao et al. (2021). In the present study, thirteen fresh rust specimens were collected on eight Rosaceae hosts, such as *Duchesnea indica*, *Potentilla freyniana*, *P. kleiniana*, *Rosa roxbunghii*, *R. laevigata*, *Rosa* sp., *Rubus coreanus* and *Ru. parrifolius* in Guizhou Province. This study aimed to determine the taxonomic status of the parasitic pecies of the Rosaceae in Guizhou Province through morphological and molecular characters. Meanwhile, we hope to contribute a significant amount of molecular data that may aid future studies and phylogenetic placement of *Phragmidium* in the Pucciniales.

## Materials and methods

### Sampling and microscopy observation

Thirteen fresh rust specimens were collected on branch and leaf from eight species of Rosaceae, *Duchesnea indica*, *Potentilla freyniana*, *P. kleiniana*, *Rosa roxbunghii*, *R. laevigata*, *Rosa* sp., *Rubus coreanus* and *R. parrifolius* in Guizhou Province, China. The spores from specimens were mounted in sterile water, on slides and observed using a Zeiss Scope 5 compound microscope (Axioscope 5, Jena, Germany), and photographed with an AxioCam 208 color (Jena, Germany) camera and saved as JPG files. Approximately 30 measurements were made of each feature using the ZEN 2.0 (blue edition) software. The Flora of China (http://www.efloras.org/flora_page.aspx?flora_id=4) was used to identify host plants (Liu et al. 2018). The rust specimens were deposited in the HGUP Herbarium of Department of Plant Pathology, Agricultural College, Guizhou University. Taxonomic details of our novel taxa were submitted to MycoBank (www.mycobank.org).

### DNA extraction, PCR and sequencing

Rust spores were scraped from fresh plant tissues using a sterile scalpel. Total DNA of rust spores was extracted with a BIOMIGA Fungus Genomic DNA Extraction Kit (GD2416) according to the manufacturer's protocol. Targeted sequences of internal transcribed spacer of rDNA (ITS) was amplified using primers ITS4rust (5'-CAGATTACAAATTTGGGCT-3') (Beenken et al. 2012) and Rust2inv (5'-GATGAAGAACACAGTGAAA-3') (Aime 2006), and the large subunit (*LSU*) of the ribosomal RNA gene was amplified using the primers No.4 (5'-ACCCGCTG AATTTAAGCATAT-3')/No.11 (5'-CTCCTTGGTCCGTGTTTCAAGACGC-3') (Van der Auwera et al. 1994), or LR6 (5'-CGCCAGTTCTGCTTACC-3') (Vilgalys and Hester 1990), and LR0R (5'-ACCCGCTGAACTTAAGC-3') (Hopple 1994). The PCR cycling conditions were as described by Liu et al (2018). The PCR amplicons from purification and sequencing were carried out at Sangon Biotech (Chengdu, China). Newly-generated sequences were deposited in GenBank (Table 1).

### Phylogenetic analyses

81 sequences, including originated from thirteen specimens and related sequences of *Phragmidium* spp. were aligned in the online version of MAFFT v. 7.307 (Katoh and Standley 2016). *Trachyspora intrusa* (BPI 843828) was selected as outgroup (Liu et al. 2020). The alignment document was edited using MEGA6 (Tamura et al. 2013) and manually adjusted when necessary.

All relevant sequences of ITS—*LSU* dataset were conducted using maximum likelihood (ML), maximum parsimony (MP) and Bayesian inference (BI) methods. ML analysis was performed using RAxML-HPC2 v.8.2.12 (Stamatakis 2014).

**Table 1.** Specimens and GenBank accession numbers of rust isolates included in this study.

| Species | Voucher specimens | Host | Locality | ITS | *LSU* |
|---|---|---|---|---|---|
| *Phragmidium andersoni* | HMAS-53231 T | *Potentilla fruticosa* | Sinkiang, China | N/A | MG669120 |
| *Ph. altaicum* | BJFCR03247 | *Rosa albertii* | China | MH285385 | MH285381 |
| | BJFCR03246 | *Rosa albertii* | China | MH285384 | MH285380 |
| | BJFCR03217 T | *Rosa albertii* | China | MH285383 | MH285379 |
| *Ph. barclayi* | HMAS-67281 | *Rubus austrotibetanus* | Tibet, China | N/A | MG669117 |
| *Ph. barnardii* | BRIP 56945 | *Rubus parvifolius* | South Africa | N/A | KT199402 |
| ***Ph. barnardii*** | **HGUP21035** | ***Rubus parvifolius*** | **Guizhou, China** | **OL684828** | **OL684839** |
| *Ph. biloculare* | BPI:881121 | *Potentilla flabellifolia* | USA | N/A | JF907670 |
| *Ph. butleri* | HMAS-67841 | *Rosa macrophylla* | Tibet, China | N/A | MG669118 |
| *Ph. chayuensis* | BJFC-R02532 T | *Rosa duplicata* | Tibet, China | N/A | MG669112 |
| | BJFC-R03014 T | *Rosa duplicata* | Tibet, China | N/A | MG669113 |
| *Ph. cibanum* | BJFCR02528 T | *Rubus niveus* | Tibet, China | MH128370 | MG669110 |
| | BJFCR03012 T | *Rubus niveus* | Tibet, China | MH128371 | MG669111 |
| ***Ph. duchesneae-indica*** | **HGUP21031** | ***Duchesnea indica*** | **Guizhou, China** | **OL684824** | **OL684835** |
| | **HGUP21032** | ***Duchesnea indica*** | **Guizhou, China** | **OL684825** | **OL684836** |
| *Ph. fragariae* | WM 1317 | *Potentilla sterilis* | Europe | N/A | AF426217 |
| *Ph. fusiforme* | T-10 | *Rosa pendulina* | Switzerland | N/A | AJ715522 |
| *Ph. fructigenum* | HMUT100472 | *Rosa glomerata* | Guangdong, China | N/A | KU059168 |
| *Ph. griseum* | BJFCR03449 | *Rubus crataegifoliu* | Beijing, China | MN264712 | MN264730 |
| | BJFCR03451 | *Rubus crataegifoliu* | Beijing, China | MN264713 | MN264731 |
| | HMAS56906 | *Rubus crataegifoliu* | Beijing, China | N/A | MG669115 |
| *Ph. handelii* | BJFC-R01030 | *Rosa webbiana* | Gansu, China | N/A | KP407631 |
| *Ph. ivesiae* | BPI-877968 | *Potentilla gracilis* | USA | N/A | JF907673 |
| | BPI-863637 | *Potentilla gracilis* | USA | N/A | JF907672 |
| | BJFC-R01421 | *Rosa webbiana* | Gansu, China | N/A | KP407628 |
| *Ph. japonicum* | HMAS41585 | *Rosa laevigata* | Fujian, China | MN264716 | MN264734 |
| | IBAR8174 | *Rosa luciae* | Ibaraki, Japan | MN882389 | MN848143 |
| *Ph. jiangxiense* | BJFCR03452 | *Rosa laevigata* | Jiangxi, China | MN264714 | MN264732 |
| | BJFCR03453 T | *Rosa laevigata* | Jiangxi, China | MN264715 | MN264733 |
| *Ph. leucoaecium* | BJFCR02116 | *Rosa* sp. | Yunnan, China | MN264718 | MN264736 |
| | BJFCR02118 T | *Rosa* sp. | Yunnan, China | MN264719 | MN264737 |
| *Ph. longissima* | BJFC-R00338 | *Rosa lichiangensis* | Yunnan, China | N/A | KP407633 |
| | BJFC-R00360 | *Rosa lichiangensis* | Yunnan, China | N/A | KP407634 |
| *Ph. mexicanum* | BPI 843961 | *Potentilla indica* | Maryland, USA | N/A | JF907660 |
| | BPI 843829 | *Potentilla indica* | Virginia, USA | N/A | JF907659 |
| *Ph. mucronatum* | RUBO | *Rosa* sp. | Bochum, Germany | N/A | KU059171 |
| | TUB 012090 | *Rosa corymbifera* | Germany | N/A | AJ715520 |
| *Ph. montivagum* | HMAS67176 | *Rosa davurica* | China | N/A | KU059173 |
| | FO 47828 | *Rosa woodsii* | NA | N/A | AF426213 |
| *Ph. octoloculare* | HMAS-140416 | *Rubus biflorus* | Tibet, China | N/A | MG669119 |
| *Ph. potentillae* | HMAS53236 | *Potentilla virgata* | Sinkiang, China | N/A | MG669114 |
| | BJFCR00961 | *Potentilla chinensis* | Qinghai, China | MN264720 | MN264738 |
| ***Ph. potentillae*** | **HGUP21034** | ***Potentilla kleiniana*** | **Guizhou, China** | **OL684827** | **OL684838** |
| *Ph. potentillae-canadensis* | BPI877886 | *Potentilla* sp. | North Carolina, USA | N/A | JF907667 |
| | BPI877885 | *Potentilla canadensis* | Maryland, USA | N/A | JF907668 |
| ***Ph. potentillae-freynianae*** | **HGUP21033 T** | ***Potentilla freyniana*** | **Guizhou, China** | **OL684826** | **OL684837** |
| *Ph. punjabense* | BA-65A T | *Rosa brunonii* | Pakistan | N/A | KX358854 |
| | BA-65B | *Rosa brunonii* | Pakistan | N/A | KX358855 |
| ***Ph. rosae-laevigatae*** | **HGUP21036 T** | ***Rosa laevigata*** | **Guizhou, China** | **OL684829** | **OL684840** |
| | **HGUP21037** | ***Rosa laevigata*** | **Guizhou, China** | **OL684830** | **OL684841** |

| Species | Voucher specimens | Host | Locality | ITS | *LSU* |
|---|---|---|---|---|---|
| *Ph. rosae-multiflorae* | HMAS71053 | *Rosa multiflora* | Shanxi, China | N/A | KU059174 |
| | HMAS94924 | *Rosa multiflora* | Zhejiang, China | N/A | KU059175 |
| | BJFCR03454 | *Rosa multiflora* | Jiangxi, China | MN264721 | MN264739 |
| ***Ph. rosae-roxburghii*** | **HGUP21025** [T] | ***Rosa roxburghii*** | **Guizhou, China** | **OL684818** | **OL684831** |
| | **HGUP21026** | ***Rosa roxburghii*** | **Guizhou, China** | **OL684819** | **OL684832** |
| | **HGUP21027** | ***Rosa roxburghii*** | **Guizhou, China** | **OL684820** | **N/A** |
| | **HGUP21028** | ***Rosa* sp.** | **Guizhou, China** | **OL684821** | **OL678103** |
| *Ph. rosae-rugosae* | BJFCR03455 | *Rosa rugosa* | Jiangxi, China | MN264722 | MN264740 |
| | BJFCR03456 | *Rosa rugosa* | Beijing, China | MN264723 | MN264741 |
| *Ph. rubi-idaei* | WM 1024 | *Rubus idaeus* | Europe | N/A | AF426215 |
| | BRIP 59372 | *Rubus idaeus* | Australia | N/A | MW147044 |
| *Ph. rubi-oldhami* | HMAS-64306 | *Rubus pungens* | Sichuan, China | N/A | MG669116 |
| ***Ph. rubi-corean*** | **HGUP21029** [T] | ***Rubus coreanus*** | **Guizhou, China** | **OL684822** | **OL684833** |
| | **HGUP21030** | ***Rubus coreanus*** | **Guizhou, China** | **OL684823** | **OL684834** |
| *Phragmidium* sp. | HMAS41561 | *Rosa multiflora* | Fujian, China | MN264717 | MN264735 |
| *Ph. sanguisorbae* | BPI 872232 | *Sanguisorba minor* | USA | N/A | JF907674 |
| | ML 957 | *Sanguisorba minor* | Europe | N/A | AF426216 |
| *Ph. tormentillae* | BPI 843392 | *Potentilla* sp. | Maryland, USA | DQ354553 | DQ354553 |
| | BPI 877888 | *Potentilla simplex* | Tennessee, USA | N/A | JF907669 |
| *Ph. tuberculatum* | BJFCR00959 | *Rosa* sp. | Qinghai, China | N/A | KP407636 |
| | BPI 877978 | *Rosa* sp. | California, USA | N/A | KJ841919 |
| | BPI 843677 | *Rosa* sp. | Argentina | N/A | KJ841921 |
| *Ph. violaceum* | MCA2782 | *Rubus* sp. | France | DQ142909 | DQ142909 |
| | BPI 871510 | *Rubus* sp. | Oregon, USA | DQ142910 | DQ142910 |
| | BJFCR03457 | *Rubus* sp. | New Zealand | MN264724 | MN264742 |
| *Ph. warburgianum* | BJFCR03458 | *Rosa bracteata* | Japan | MN264726 | MN264744 |
| | BJFCR03459 | *Rosa bracteata* | Japan | MN264727 | MN264745 |
| *Ph. zangdongii* | BJFCR02447 [T] | *Rosa tibetica* | Tibet, China | MH128372 | MG669108 |
| | BJFCR03013 [T] | *Rosa tibetica* | Tibet, China | MH128373 | MG669109 |
| *Ph. zhouquensis* | BJFCR01516 [T] | *Rosa omeiensis* | Yunnan, China | MN264728 | MN264746 |
| | BJFCR01529 [T] | *Rosa omeiensis* | Yunnan, China | MN264729 | MN264747 |
| *Trachyspora intrusa* | BPI 843828 | *Alchemilla vulgaris* | Switzerland | DQ354550 | |

[T] = Type specimens. New specimens are in bold typeface.

Gaps were treated as "missing". The MP analysis of the two loci (ITS and *LSU*) was implemented with PAUP v. 4.0b10 (Swofford 2002). The phylogenetic trees were generated using the heuristic search option with tree bisection reconnection (TBR) branch swapping and 1000 random sequence additions. The maxtrees was set to 5000. The tree length (TL), consistency index (CI), homoplasy index (HI), retention index (RI), and rescaled consistency index (RC) were calculated. Bayesian inference analysis was inferred by MrBayes 3.2.6 (Ronquist et al. 2012). The best model for two loci (ITS and *LSU*) was determined by MrModeltest v2 (Nylander 2004), ITS: HKY+G, *LSU*: GTR+I+G. BI were performed by six Markov chain Monte Carlo. These chains were run for 5 million generations, sampling tree every 100 generations. The first 25% of resulting trees were discarded as burn-in phase of each analysis, and trees were saved every 5000 generations. Alignment matrices have been uploaded as an attachment.

# Results

## Phylogenetic analyses

The phylogenetic trees accommodated 82 sequences listed in Table 1. The combined alignment including ITS (493 bp) and *LSU* (544 bp) regions consisted of 1067 characters, of which 585 were constant, 89 variable characters were parsimony uninformative, and 363 were parsimony informative. We built three phylogenetic trees, ML tree, MP tree and BI tree. The MP tree was selected to represent the phylogenetic relationship of different *Phragmidium* taxa (Fig. 1). MP analysis produced the following parameters: tree length (TL) = 1011; consistency index (CI) = 0.643; homoplasy index (HI) = 0.356; retention index (RI) = 0.898; and rescaled consistency index (RC) = 0.578. *Phragmidium rubi-coreani* on *Rubus coreanus* with telial, aecial and uredinial stages formed a small branch only. *Phragmidium potentillae-freynianae* and *Ph. duchesneae-indica* constituted a distinct subclade with high statistical support (100 ML/99 MP/1.00 PP). *Phragmidium rosae-laevigatae* was phylogenetically sister to *Ph. leucoaecium*, *Ph. japonicum*, *Ph. jiangxiense* and *Phragmidium* sp. with high support (100 ML/100 MP/1.00 PP). The four aecial-uredinial fungi on *Ro. roxburghii* kept identical base composition on ITS and *LSU* gene regions and made up a distinct subclade to *Ph. warburgianum* with high support (100 ML/99 MP/1.00 PP). Our strains represented four novel taxa, which was also supported by comparison of the DNA base pair differences between our strains and related taxa on ITS and *LSU* gene region.

The hosts of the *Phragmidium* species were mainly concentrated in *Rosa*, *Rubus* and *Potentilla* of Rosaceae (Fig. 1). Eighty-one *Phragmidium* strains clustered together as a clade, which was roughly divided into three subclades (Subclade I, Subclade II and Subclade III). For Subclade I with 16 species (*Ph. rubi-coreani*, *Ph. barclayi*, *Ph. cibanum*, *Ph. violaceum*, *Ph. barnardii*, *Ph. griseum*, *Ph. rubi-idaei*, *Ph. altaicum*, *Ph. tuberculatum*, *Ph. octoloculare*, *Ph. sanguisorbae*, *Ph. punjabense*, *Ph. rubi-oldhami*, *Ph. butleri*, *Ph. zhouquensis* and *Ph. fragariae*) (67 ML/59 MP), their hosts belonged to *Rosa*, *Rubus*, *Potentilla*, and *Sanguisorba*. *Phragmidium rubi-coreani* and *Ph. rubi-ideai* associated with host plants on the generic level had obvious genetic distance. Subclade II included 18 *Phragmidium* taxa (*Ph. biloculare*, *Ph. potentillae*, *Ph. ivesiae*, *Ph. montivagum*, *Ph. fructigenum*, *Ph. zangdongii*, *Ph. fusiforme*, *Ph. handelii*, *Ph. rosae-rugosae*, *Ph. mucronatum*, *Ph. chayuensis*, *Ph. longissima*, *Ph. rosae-multiflorae*, *Ph. mexicanum*, *Ph. potentillae-canadensis*, *Ph. potentillae-freynianae*, *Ph. duchesneae-indica* and *Ph. tormentillae*) (95 ML), but their host plants only referred to *Rosa*, *Potentilla* and *Duchesnea*. *Phragmidium potentillae-freynianae* and *Ph. duchesneae-indica* belonging to different generic host plants were accommodated to a branch (100 ML/99 MP/1.00 PP), but *Ph. mexicanum* and *Ph. potentillae-canadensis* formed a clade (99 ML/86 MP/1.00 PP) separated from *Ph. potentillae-freynianae* with the congeneric host plants. *Phragmidium tormentillae* associated with *Potentilla canadensis* (*P. simplex*) as its host formed an independent branch (97 ML/61 MP/0.94 PP). The *Phragmidium* host plants in Subclade III (*Ph. rosae-roxburghii*, *Ph. warburgianum*, *Ph. japonicum*,

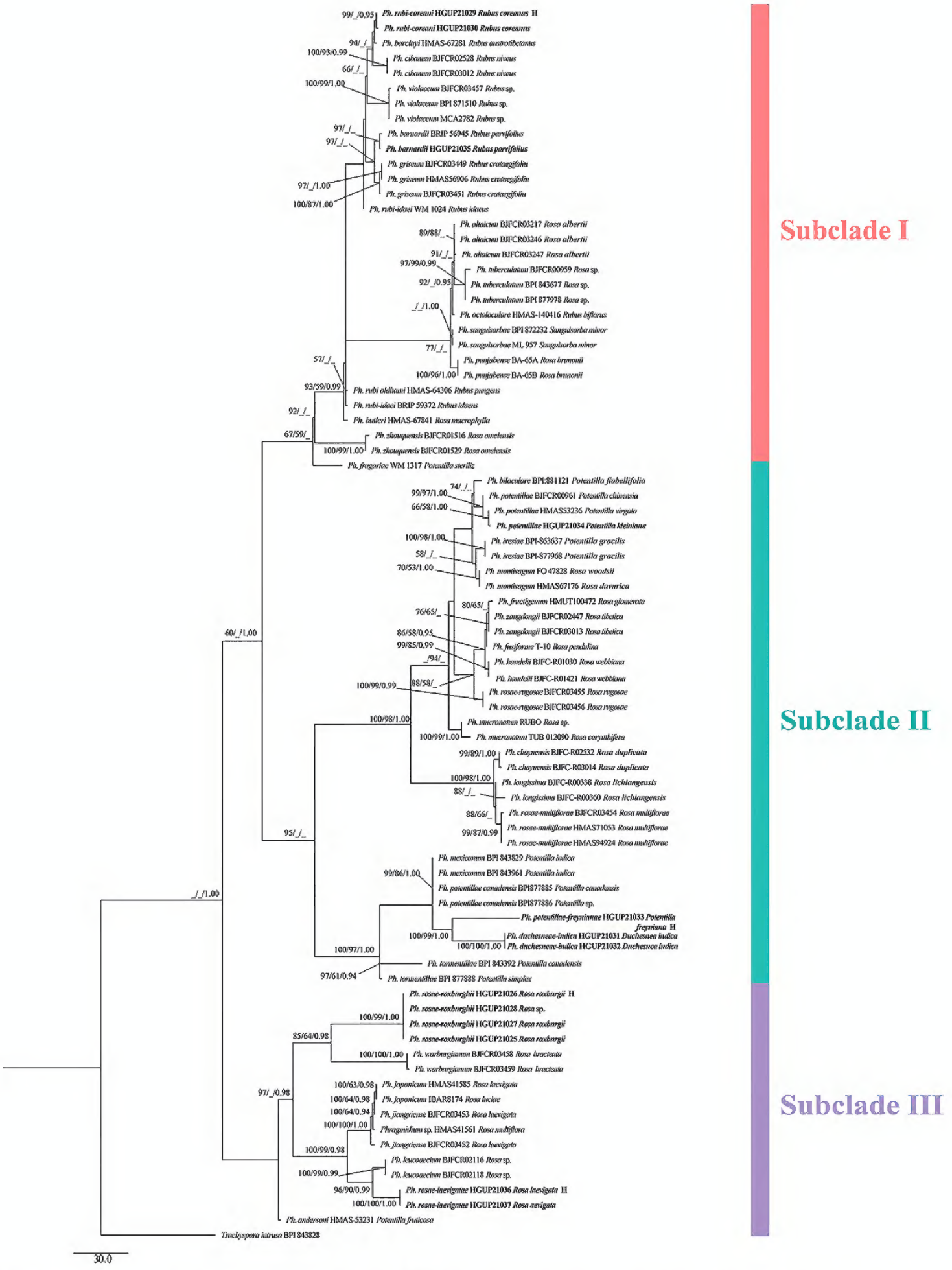
Ph. rubi-coreani HGUP21029 Rubus coreanus H
Ph. rubi-coreani HGUP21030 Rubus coreanus
Ph. barcleyi HMAS-67281 Rubus austrotibetanus
Ph. cibanum BJFCR02528 Rubus niveus
Ph. cibanum BJFCR03012 Rubus niveus
Ph. violaceum BJFCR03457 Rubus sp.
Ph. violaceum BPI 871510 Rubus sp.
Ph. violaceum MCA2782 Rubus sp.
Ph. barnardii BRIP 56945 Rubus parvifolius
Ph. barnardii HGUP21035 Rubus parvifolius
Ph. griseum BJFCR03449 Rubus crataegifolia
Ph. griseum HMAS56906 Rubus crataegifolia
Ph. griseum BJFCR03451 Rubus crataegifolia
Ph. rubi-idaei WM 1024 Rubus idaeus
Ph. altaicum BJFCR03217 Rosa albertii
Ph. altaicum BJFCR03246 Rosa albertii
Ph. altaicum BJFCR03247 Rosa albertii
Ph. tuberculatum BJFCR00959 Rosa sp.
Ph. tuberculatum BPI 843677 Rosa sp.
Ph. tuberculatum BPI 877978 Rosa sp.
Ph. octoloculare HMAS-140416 Rubus biflorus
Ph. sanguisorbae BPI 872232 Sanguisorba minor
Ph. sanguisorbae ML 957 Sanguisorba minor
Ph. punjabense BA-65A Rosa brunonii
Ph. punjabense BA-65B Rosa brunonii
Ph. rubi okliani HMAS-64306 Rubus pungens
Ph. rubi-idaei BRIP 59372 Rubus idaeus
Ph. butleri HMAS-67841 Rosa macrophylla
Ph. zhouquensis BJFCR01516 Rosa omeiensis
Ph. zhouquensis BJFCR01529 Rosa omeiensis
Ph. fragariae WM 1317 Potentilla sterilis
Ph. biloculare BPI:881121 Potentilla flabellifolia
Ph. potentillae BJFCR00961 Potentilla chinensis
Ph. potentillae HMAS53236 Potentilla virgata
Ph. potentillae HGUP21034 Potentilla kleiniana
Ph. ivesiae BPI-863637 Potentilla gracilis
Ph. ivesiae BPI-877968 Potentilla gracilis
Ph. montivagum FO 47828 Rosa woodsii
Ph. montivagum HMAS67176 Rosa davurica
Ph. fructigenum HMUT100472 Rosa glomerata
Ph. zangdongii BJFCR02447 Rosa tibetica
Ph. zangdongii BJFCR03013 Rosa tibetica
Ph. fusiforme T-10 Rosa pendulina
Ph. handelii BJFC-R01030 Rosa webbiana
Ph. handelii BJFC-R01421 Rosa webbiana
Ph. rosae-rugosae BJFCR03455 Rosa rugosae
Ph. rosae-rugosae BJFCR03456 Rosa rugosae
Ph. mucronatum RUBO Rosa sp.
Ph. mucronatum TUB 012090 Rosa corymbifera
Ph. chayuensis BJFC-R02532 Rosa duplicata
Ph. chayuensis BJFC-R03014 Rosa duplicata
Ph. longissima BJFC-R00338 Rosa lichiangensis
Ph. longissima BJFC-R00360 Rosa lichiangensis
Ph. rosae-multiflorae BJFCR03454 Rosa multiflorae
Ph. rosae-multiflorae HMAS71053 Rosa multiflorae
Ph. rosae-multiflorae HMAS94924 Rosa multiflorae
Ph. mexicanum BPI 843829 Potentilla indica
Ph. mexicanum BPI 843961 Potentilla indica
Ph. potentillae canadensis BPI877885 Potentilla canadensis
Ph. potentillae canadensis BPI877886 Potentilla sp.
Ph. potentillae-freynianae HGUP21033 Potentilla freyniana H
Ph. duchesneae-indica HGUP21031 Duchesnea indica
Ph. duchesneae-indica HGUP21032 Duchesnea indica
Ph. tormentillae BPI 843392 Potentilla canadensis
Ph. tormentillae BPI 877888 Potentilla simplex
Ph. rosae-roxburghii HGUP21026 Rosa roxburgii H
Ph. rosae-roxburghii HGUP21028 Rosa sp.
Ph. rosae-roxburghii HGUP21027 Rosa roxburgii
Ph. rosae-roxburghii HGUP21025 Rosa roxburgii
Ph. warburgianum BJFCR03458 Rosa bracteata
Ph. warburgianum BJFCR03459 Rosa bracteata
Ph. japonicum HMAS41585 Rosa laevigata
Ph. japonicum IBAR8174 Rosa luciae
Ph. jiangxiense BJFCR03453 Rosa laevigata
Phragmidium sp. HMAS41561 Rosa multiflora
Ph. jiangxiense BJFCR03452 Rosa laevigata
Ph. leucoaecium BJFCR02116 Rosa sp.
Ph. leucoaecium BJFCR02118 Rosa sp.
Ph. rosae-laevigatae HGUP21036 Rosa laevigata H
Ph. rosae-laevigatae HGUP21037 Rosa aevigata
Ph. andersonii HMAS-53231 Potentilla fruticosa
Trachyspora intrusa BPI 843828
30.0
Subclade I
Subclade II
Subclade III

**Figure 1.** The maximum parsimony tree of 42 *Phragmidium* taxa based on ITS and *LSU* genes; host plants are also given.

*Ph. jiangxiense*, *Phragmidium* sp., *Ph. leucoaecium*, *Ph. rosae-laevigatae*, *Ph. andersoni*) belonged to *Rosa* and *Potentilla*. *Phragmidium rosae-laevigatae* and *Ph. rosae-roxburghii* with the same generic host plants did not group together (97 ML /0.98 PP).

*Phragmidium japonicum*, *Ph. jiangxiense* and *Phragmidium* sp. (HMAS51561) all from *Rosa* formed a branch (100 ML/100 MP/1.00 PP). *Phragmidium andersoni* collected from *Potentilla fruticosa* formed an independent branch.

RA×ML and MP bootstrap support values (MP ≥ 50%), and Bayesian posterior probability (PP ≥ 0.90) are marked on the nodes as (ML/MP/PP). Specimens from current study have put in bold and put an H in the selected holotypes. The outgroup was *Trachyspora intrusa* (BPI 843828). The scale bar indicates 30 expected changes per site.

## Taxonomy

***Phragmidium rosae-roxburghii* J.E. Sun & Yong Wang bis, sp. nov.**
MycoBank No: MB845041
Figs 2, 3

**Diagnosis.** *Phragmidium rosae-roxburghii* easily to be distinguished by its unique square to diamond-shaped urediniospores.

**Holotype.** China. Guizhou Province, Panzhou city, 25°89'61"N, 104°56'07"W, 750 m, 21 Mar 2021, on *Rosa roxburghii*, coll. J.E. Sun & Y.Q. Yang, HGUP21025, ITS: OL684818, LSU: OL684831.

**Etymology.** Referring to the host, *Rosa roxburghii*, on which the fungus was first found.

**Description.** ***Spermogonia*:** unknown. ***Aecia*** formed on gold distinct, circular lesions on both sides of the stems, petioles and leaves, rarely produced on the abaxial leaf surface, scattered, flat oval to subglobose, powdery, 1.0–5.0 mm diam. Aeciospores formed in basipetal succession, oval o subglobose, 22–30 × 14–22 µm (mean 26 × 18 µm, n = 30), inclusions golden, to bright-yellow; wall 1.8–3.1 µm thick, colorless, mostly with irregularly elongated verrucae on the surface. ***Uredinia*** produced on the abaxial leaf surface, scattered to gregarious, hypophyllous, orange-colored or white, powdery, oval to rounded, 0.1–1.0 mm diam, paraphysis in the periphery of the uredinia, curved, 30–55 × 9–20 µm, colorless thin-walled. Urediniospores generally angular, square to diamond-shaped, yellowish to orange-colored, 20–30 × 16–21 µm (mean: 25 × 19 µm, n = 30), thick-walled, 0.5–2.0 µm thick, colorless, regularly echinulate with stout spines.

Rust diseases symptoms: In the early stage (March) of rust disease yellowish-orange powdery aecia formed on the stems and petioles on *Rosa roxburghii* and *Rosa* sp., the aecia were scattered, flat oval or nearly round and bordered (Fig. 2). In middle of June (Fig. 3), the upper surface of the lower leaves was turning yellow and orange spots gradually appeared on the under surface caused by uredinia, which are powdery, aggregated but without obvious boundaries.

**Habitat.** *Rosa roxburghii*, *Rosa* sp.

**Known distribution.** China, Guizhou Province.

**Additional material examined.** China. Guizhou Province: Duyun city, 26°45'88"N, 106°98'42"W, 820 m, 22 Jun 2021, on *Rosa roxburghii*, coll. J.E. Sun,

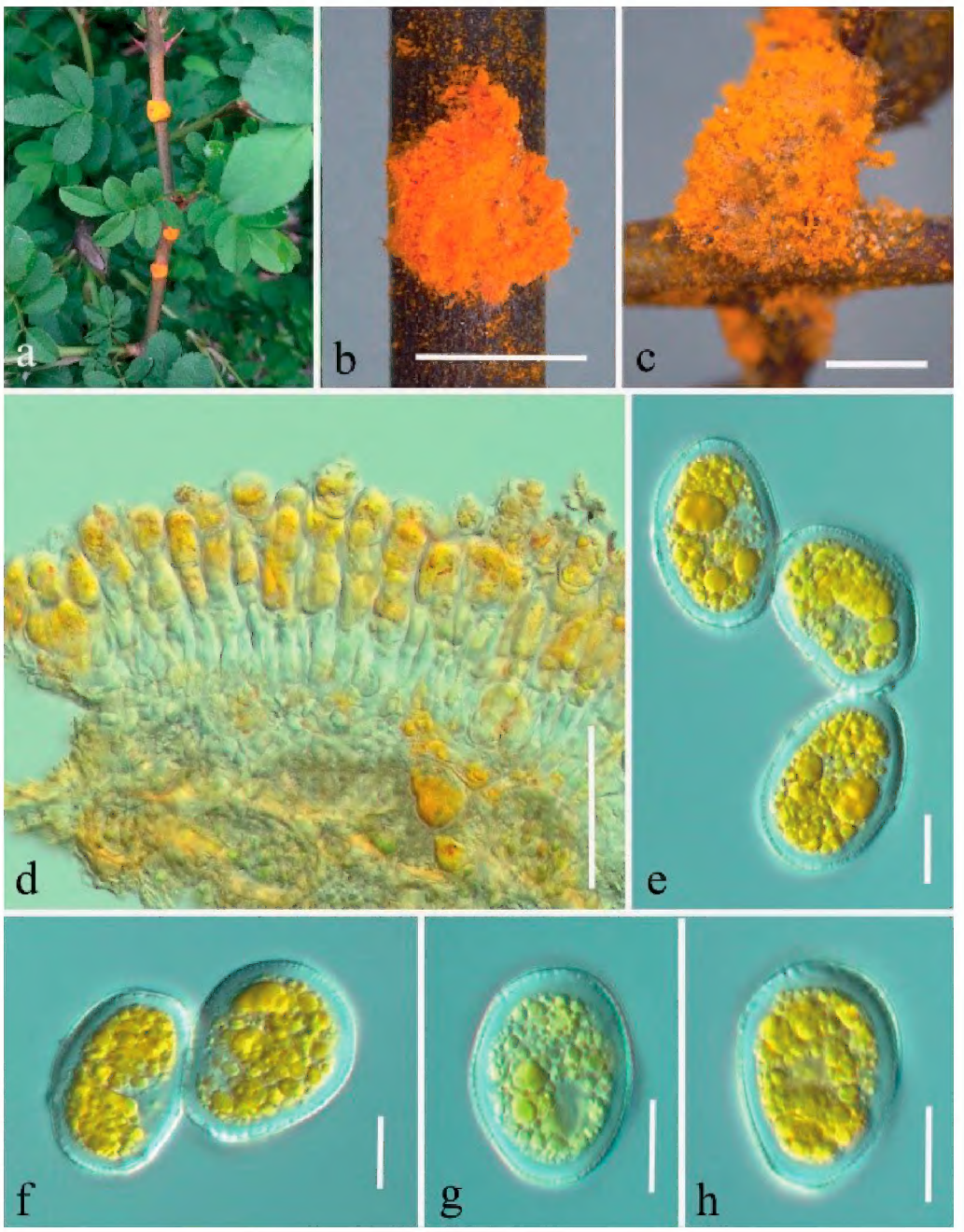


**Figure 2.** *Phragmidium rosae-roxburghii* sp. nov. (HGUP21025, holotype) on *Rosa roxburghii* **a–c** aecia on stem and leaf pieces. **d** longitudinal section of aecium **e–h** aeciospores. Scale bars: 2 mm (**b–c**); 50 μm (**d**); 10 μm (**e–h**).

HGUP21026; Tongren city, 28°14'09"N, 108°34'03"W, 810 m, 04 Sep 2021, on *Rosa roxburghii*, coll. J.E. Sun, HGUP21027; Guiyang city, 26°44'74"N, 106°58'67"W, 960 m, 27 Mar, 2021, on *Rosa* sp., coll. J.E. Sun, HGUP21028.

**Notes.** *Phragmidium rosae-roxburghii* was the first species of *Phragmidium* described on *Rosa roxburghii*. It is easily to distinguish species by its unique square to diamond-shaped urediniospores, since in other *Phragmidium* species the urediniosporas are oval to nearly spherical (Yun et al. 2011; Ono 2012; Zhuang et al. 2012; Yang et al. 2015; Liu et al. 2018, 2019, 2020; Ono and Wahyuno 2019). In phylogeny, this species only kept a close relationship to *Ph. warburgiana* (Fig. 1) but its urediniospores are yellowish to orange-colored different to *Ph. warburgiana* with colorless urediniospores (Ono 2012). We proposed *Ph. rosae-roxburghii* as a new taxon.

**Figure 3.** *Phragmidium rosae-roxburghii* sp. nov. (HGUP21026) on *Rosa roxburghii* **a** appearance of infected plants **b** uredinia on a leaf **c** longitudinal section of uredinium **d** paraphyses **e–i** urediniospores. Scale bars: 5 mm (**b**); 50 μm (**c**); 25 μm (**d**); 12.5 μm (**e–i**).

***Phragmidium rubi-coreani*** **J.E. Sun & Yong Wang bis, sp. nov.**
MycoBank No: MB845042
Fig. 4

**Diagnosis.** *Phragmidium rubi-coreani* differs to *Ph. barclayi* by teliospores with fewer cells and shorter pedicels.

**Holotype.** China. Guizhou Province: Guiyang city, 26°45'86"N, 106°98'77"W, 970 m, 11 Apr, 2021, on *Rubus coreanus*, coll. J.E. Sun, HGUP21029, ITS: OL684822, LSU: OL684833.

**Etymology.** Referring to the host, *Rubus coreanus*, on which this species grows.

**Description.** ***Spermogonia*****:** unknown. ***Aecia*** golden, produced on the abaxial leaf surface, hypophyllous, and 2.5–3.5 mm diam, subglobose to globose, powdery, 2.5–3.5 mm diam. Aeciospores produced in basipetal succession, subglobose, 14–24 × 10–23 μm (mean 19 × 16 μm, n = 30), bright yellow contents, thick-walled, 1.0–4.0 μm, colorless, echinulate; paraphyses clavate, not or weakly incurved, 38–61 μm long, thick-walled, wall 2.0–2.5 μm thick. ***Telia*** hypophyllous, scattered, 0.3–0.5 mm diam, chocolate-brown. Teliospores ellipsoid to cylindrical, 3–5 celled, constricted at

the septa, bright orange, chocolate-brown to gray-brown, 29–74 × 14–37 µm (mean 50 × 25 µm, n = 30), thick-walled, wall 1.8–3.5 µm thick, colorless to chocolate-brown; pedicels not swollen at the base, 8–34 µm long, colorless. ***Uredinia*** formed on circular lesions on both sides of the leaves, powdery, yellow distinct, hypophyllous scattered, nearly oval, surrounded by host epidermis, 0.5–1.0 mm diam. Urediniospores: uredo-type, subglobose to oval, produced in basipetal succession, golden, or bright-yellow, 19–27 × 15–25 µm (mean 23 × 20 µm, n = 30), thick-walled, wall 0.8–1.5 µm thick, colorless, densely and minutely echinulate.

Rust diseases symptoms: The golden and powdery aecia were first produced on the underside of leaves. Then, scattered uredinia were formed, orange-colored and forming small round spots on the leaves. Chocolate-brown telia were produced on the leaf remnants (Fig. 4).

**Habitat.** *Rubus coreanus*.

**Known distribution.** China, Guizhou Province.

**Figure 4.** *Phragmidium rubi-coreani* sp. nov. (HGUP21029, holotype) on *Rubus coreanus* **a** gross features of infected leaves **b** uredinia on a leaf **c–d** longitudinal section of uredinium **e** paraphyses **f** urediniospores **g** aecia on a leaf **h** longitudinal section of aecium **i–j** aeciospores **k** telia on a leaf **l** longitudinal section of telium **m–n** Teliospores. Scale bars: 2 mm (**b**); 1 mm (**g, k**); 50 µm (**c–e, h, l**); 10 µm (**f**); 25 µm (**i–j, m–n**).

**Additional material examined.** China. Guizhou Province: Guiyang city, 27°10'30"N, 106°99'91"W, 830 m, 09 Apr 2021, on *Rubus coreanus*, coll. J.E. Sun, HGUP21030.

**Notes.** In the phylogenetic tree, *Phragmidium rubi-coreani*, *Ph. barclayi* and *Ph. cibanum* formed a branch (Fig. 1). However in morphology, teliospores of *Phragmidium rubi-coreani* have fewer septa and shorter pedicels (3–5-celled, 8–34 μm long) than *Ph. barclayi* (5–8-celled, 60–150 μm long) and *Ph. cibanum* (5–7-celled, 70–108 μm long) (Liu et al. 2018). Meanwhile, most reported *Phragmidium* taxa produce longer teliospores, such as *Ph. zangdongii* (29–74 × 14–37 μm vs. 82–110 × 23–31 μm); *Ph. kanas* (29–74 × 14–37 μm vs. 134–198 × 19–31 μm); *Ph. potentillae-canadensis* (29–74 × 14–37 μm vs. 48.1–86.8 × 30.1–33.3 μm) than the present species (Yun et al. 2011; Liu et al. 2018; Zhao et al. 2021). Thus, our fungus represented a novel taxon.

***Phragmidium potentillae-freynianae*** **J.E. Sun & Yong Wang bis, sp. nov.**
MycoBank No: MB845043
Fig. 5

**Diagnosis.** Different from the related taxa by its urediniospores catenulate, such as *Ph. chayuensis*, *Ph. cibanum* and *Ph. tormentillae*.

**Holotype.** China. Guizhou Province;, Guiyang city, 26°44'70"N, 106°59'65"W, 801 m, 27 Mar 2021, on *Potentilla freyniana*, coll. J.E. Sun, HGUP21033, ITS: OL684826, LSU: OL684837.

**Etymology.** Referring to the host, *Potentilla freyniana*, on which the fungus was first found.

**Description.** ***Spermogonia***, ***aecia*** and ***telia*** not observed. ***Uredinia*** produced on the abaxial leaf surface, covering the entire lower surface of the leaves, hypophyllous, nearly oval, powdery, not surrounded by host epidermis, 0.1–1.0 mm diam, on densely orange spot, 0.1–1.0 mm diam. Urediniospores: uredo-type, subglobose to oval, produced in basipetal succession, 19–24 × 18–24 μm (mean 21.5 × 21 μm, n = 30), golden, or bright-yellow; thin-walled, wall 0.4–1.4 μm thick, colorless, densely and minutely echinulate.

Rust diseases symptoms: Large areas of orange powdery uredinia, covering almost the entire lower surface of the leaves, which are aggregated but without obvious boundaries (Fig. 5).

**Habitat.** *Potentilla freyniana*.

**Known distribution.** China, Guizhou Province.

**Notes.** In the phylogenetic tree, *Phragmidium potentillae-freynianae* formed a well-supported clade allied to *Ph. duchesneae-indica* (Fig. 1). Morphologically, its urediniospores are bigger than *Ph. duchesneae-indica* (21.5 × 21 μm vs. 13–19 × 11–17 μm) (Zhao et al. 2021). The comparison of DNA base composition supports the morphological separation of this fungus as a new species.

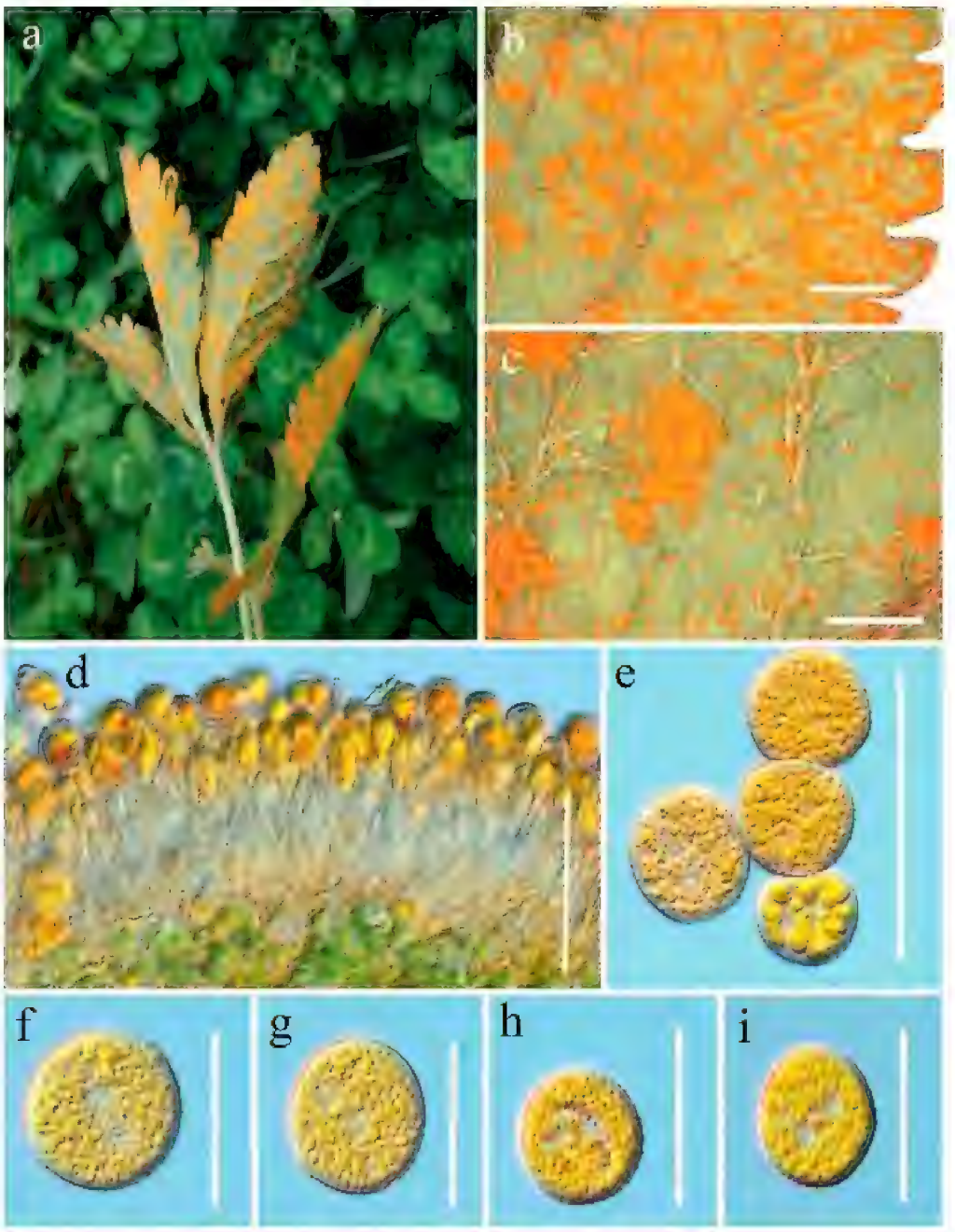


**Figure 5.** *Phragmidium potentillae-freynianae* sp. nov. (HGUP21033, holotype) on *Potentilla freyniana*. **a–c** uredinia on leaves **d** longitudinal section of uredinium **e–i** urediniospores. Scale bars: 2 mm (**b–c**); 50 μm (**d–e**); 25 μm (**f–i**).

***Phragmidium rosae-laevigatae*** **J.E. Sun & Yong Wang bis, sp. nov.**
MycoBank No: MB845044
Fig. 6

**Diagnosis.** Different from *Ph. Jiangxiense* mainly because of bigger urediniospores.

**Holotype.** China. Guizhou Province: Panzhou city, 25°64'56"N, 104°84'35"W, 1800 m, 19 Jul 2021, on *Rosa laevigata*, coll. J.E. Sun, HGUP21036, ITS: OL684829, LSU: OL684840.

**Etymology.** Referring to the host, *Rosa laevigata*, on which the fungus was first found.

**Description.** ***Spermogonia*** and ***aecia*** not observed. ***Uredinia*** produced on the abaxial leaf surface, hypophyllous, subglobose to globose, powdery, 0.1–0.5 mm diam, yellow, peripherally parphyses, hyaline, 20–31 × 10–17 μm. Urediniospores square to diamond-shaped, oval to nearly spherical, 23–35 × 16–30 μm (mean 29 × 23 μm, n = 30), orange-

colored, thick-walled 0.5–2.0 μm thick, colorless, regularly echinulate with stout spines on the surface. ***Telia*** scattered compact, hypophyllous, golden, 0.1–0.5 mm diam. Teliospores (immature) oval, 24–60 × 8–20 μm (mean 50.5 × 25.5 μm, n = 30), with apical papillae (4.0–7.0 μm high, n = 10), too immature to know how many cells, orange-yellow; pedicels swollen at the base, 15–26 μm long, colorless, disconnected easily; wall 0.5–2.0 μm thick.

Rust diseases symptoms: As shown in Fig. 6, Uredinia and telia, which are bright-yellow and powdery are produced almost simultaneously on the lower surface of the yellowing and wilting leaves.

**Habitat.** *Rosa laevigata*.

**Known distribution.** China, Guizhou Province.

**Additional material examined.** China. Guizhou Province: Panzhou city, 25°61'81"N, 104°83'61"W, 1790 m, 19 Jul 2021, on *Rosa laevigata*, coll. J.E. Sun, HGUP21037.

**Notes.** Phylogenetically, *Phragmidium rosae-laevigatae* kept a close relationship to *Ph. leucoaecium*, *Ph. japonicum* and *Ph. jiangxiense* (Fig. 1). Morphologically, *Phragmidium rosae-laevigatae* has bigger urediniospores than *Ph. jiangxiense* (23–35 × 16–30 μm vs. 15–23 × 11–18 μm), but the uredinia and urediniospores of

**Figure 6.** *Phragmidium rosae-laevigatae* sp. nov. (HGUP21036, holotype) on *Rosa laevigata* **a** gross features of infected leaves **b** uredinia and telia on a leaf **c** longitudinal section of telium **d** immature teliospores **e** longitudinal section of uredinium **f–h** urediniospores. Scale bars: 1 mm (**b**); 50 μm (**c, e**); 12.5 μm (**d, f–h**).

*Ph. leucoaecium* and *Ph. japonicum* were not observed (Liu et al. 2020). The comparison of DNA base composition also supported morphological conclusion. Thus, this fungus was also introduced as one novel taxon herein.

***Phragmidium duchesneae-indicae*** **P. Zhao & L. Cai, Fungal Diversity 5:1–58, 2021**
MycoBank No: MB557609
Fig. 7

**Description.** ***Spermogonia***, ***aecia*** and ***telia*** not observed. ***Uredinia*** produced on the abaxial leaf surface, hypophyllous, nearly oval, golden, densely bright orange-yellow, powdery, not surrounding by host epidermis, 0.3–1.2 mm diam, without paraphyses. Urediniospores produced in basipetal succession, mostly globose, 17–22 × 15–20 μm (mean 19.5 × 17.5 μm, n = 30), inclusions yellowish, or bright-yellow; thick-walled, wall 0.7–1.8 μm thick, colorless, densely and minutely echinulate. Telia and teliospores see Zhao et al (2021).

**Habitat.** *Duchesnea indica*

**Known distribution.** China, Guizhou Province.

**Material examined.** China. Guizhou Province: Guiyang city, 27°10'30"N, 106°99'91"W, 820 m, 09 Apr 2021, on *Duchesnea indica*, coll. J.E. Sun, HGUP21031;

**Figure 7.** *Phragmidium duchesneae-indica* (HGUP21031) on *Duchesnea indica* **a–c** uredinia on leaves **d** longitudinal section of uredinium **e–g** urediniospores. Scale bars: 2 mm (**b**); 1 mm (**c**); 50 μm (**d**); 12.5 μm (**e–g**).

Guiyang city, 27°09'26"N, 106°98'90"W, 734 m, 04 Sep 2021, on *Duchesnea indica*, coll. J.E. Sun, HGUP21032.

**Notes.** *Phragmidium duchesneae-indica* was first reported on *D. indica* by Zhao et al (2021). Our specimen had similar morphology to that described by Zhao et al (2021). GenBank accession numbers (ITS and *LSU*) of *Ph. duchesneae-indicae* have not been released, and our identification is based only on a morphological comparison.

***Phragmidium potentillae* (Pers.) P. Karst., Bidrag till Kännedom av Finlands Naturoch Folk, 31: 49, 1879**

MycoBank No: MB206190

Fig. 8

**Description.** ***Spermogonia*** and ***aecia*** not observed. ***Uredinia*** produced on the abaxial leaf surface, hypophyllous, nearly oval, powdery, densely bright orange, nearly oval, surrounding by host epidermis, 0.8–1.5 × 0.4–0.7 mm, and densely bright orange. Urediniospores angular to squarish, oval to nearly globose, produced in basipetal succession, 17–26 × 14–22 μm (mean 21.5 × 18 μm, n = 30), or bright–yellow to orange, immature urediniospores are colorless; thick-walled, wall 0.6–1.3

**Figure 8.** *Phragmidium potentillae* (HGUP21034) on *Potentilla kleiniana* **a–c** uredinia on leaves **d** longitudinal section of uredinium **e–j** urediniospores. Scale bars: 1 mm (**c**); 50 μm (**d**); 12.5 μm (**e–j**).

μm thick, colorless, densely and minutely echinulate. Telia and teliospores see Liu et al (2018).

**Habitat.** *Potentilla kleiniana*

**Known distribution.** China: Guizhou Province, Qinghai Province, Sinkiang Province; USA, the United Kingdom, Australia, Tasmania and Japan.

**Material examined.** China. Guizhou Province: Guiyang city, 27°09'26"N, 106°98'90"W, 730 m, 22 Jun 2021, on *Potentilla kleiniana*, coll. J.E. Sun, HGUP21034.

**Notes.** In the phylogenetic tree, HGUP21034 clustered with two sequences of specimens of *Phragmidium potentillae* (Fig. 1). The uredinia of *P. potentillae* described by Liu et al (2018), as 0.2–0.8 mm diam, smaller than in the specimen examined, 0.8–1.5 × 0.4–0.7 mm, the urediniospores mostly globose and echinulate, (18–25 × 15–21 μm vs. 17–26 × 14–22 μm).

***Phragmidium barnardii*** **Plowr. & G. Winter, Revue Mycologique Toulouse 8 (32): 208 (1886)**

MycoBank No: MB249450

Fig. 9

**Description.** ***Spermogonia***, ***aecia*** and ***telia*** not observed. Uredinia produced on the abaxial leaf surface, hypophyllous, scattered to gregarious, oval to globose, orange,

**Figure 9.** *Phragmidium barnardii* (HGUP21035) on *Rubus* sp. **a–d** uredinia on leaves **e** longitudinal section of uredinium **f–h** urediniospores. Scale bars: 1 mm (**d**); 50 μm (**e**); 12.5 μm (**f–h**).

powdery, 0.1–1.0 mm diam, with hyaline and curved paraphyses, 26–39 × 10–13 µm. Urediniospores orange, 16–19 × 15–18 µm (mean: 17.5 × 16.5 µm, n = 30), nearly globose; thick-walled 1.3–2.2 µm, colorless, regularly echinulate with stout spines.

**Habitat.** *Rubus* sp.

**Known distribution.** China, Guizhou Province; South Africa.

**Material examined.** China. Guizhou Province: Duyun city, 27°26'05"N, 107°38'91"W, 870 m, 26 Jun 2021, on *Rubus* sp., coll. J.E. Sun, HGUP21035.

**Notes:** *Phragmidium barnardii* was first reported on *Rubus* sp. by Winter (1886). Its DNA data was established by McTaggart et al (2016), although without description of morphological characteristics. We confirmed the specimens (HGUP21035) as *Ph. barnardii*, through phylogenetic analyse with DNA data from McTaggart et al. (2016).

## Discussion

More than 70 *Phragmidium* species have been described in China, while many species without molecular data (Cummins 1931; Arthur 1934; Wahyuno et al. 2001; Cummins and Hiratsuka 2003; Zhuang et al. 2012; Yang et al. 2015; Ali et al. 2017). Recently, morphology and molecular data were gradually combined and used to describe the diversity of species in *Phragmidium* (Liu et al. 2018, 2019, 2020; Zhao et al. 2021). In the study, the four novel and three known species of *Phragmidium* were delineated based on phylogeny of the ITS and *LSU* gene regions and on morphological features.

The host plants of *Ph. punjabense*, *Ph. warburgianum*, *Ph. rosae-rugosae*, *Ph. rosae-laevigatae* and *Ph. rosae-roxburghii* all belong to *Rosa*, but *Ph. potentillae-freynianae* and *Ph. potentilla* occur on *Potentilla* sp. while *Ph. rubi-coreani* and *Ph. barnardii* occur on *Rubus* sp. However, the hosts of species with close phylogenetic relationships were not necessarily in the same genus. *Phragmidium potentilla* can be found on three plants (*P. chinensia*, *P. kleiniana* and *P. virgata*), and *Ph. rosae-roxburghii* can be parasitic on two *Rosa* plants (*Rosa roxburghii* and *Rosa* sp.). It might mean that host jumps also shaped the diversity of *Phragmidium*, like Pucciniales (McTaggart et al. 2016).

*Phragmidium leucoaecium* (BJFCR02118 and BJFCR02116), *Ph. japonicum* (HMAS41585), *Ph. jiangxiense* (BJFCR03452 and BJFCR03453) and *Ph. rosae-laevigatae* (HGUP21036 and HGUP21037) from *Rosa* formed a phylogenetic lineage, while three of the latter from the same host (*Rosa laevigata*) (Liu et al. 2020). This may be explained by geographical distribution, geography, climate, etc., but contradicts the concept of obligatory parasitism. We could guess that their hosts might not reflect taxonomic status of *Phragmidium*. Interestingly, *Phragmidium tibeticum*, *Ph. sikangense* and *Ph. shensianum* were named according to the collection locations (Dai 1979; Chen 2009). Their nomenclatures contradict the concept of obligatory parasitism for rust fungi, although might be easy to be understanding.

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